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Original Article

Effect of Ammonium Sulphate Fortification on Growth Performance, Nutrient Digestibility and Nitrogen Balance of West African Dwarf Rams

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Abstract

Ammonium sulphate $(NH_4)_2SO_4$ is potential source of readily available nitrogen and sulphur which would fill the gap in feed availability to ruminants during the extended annual dry season. Hence, the use of ammonium sulphate becomes a viable alternative to growth-promoting antibiotics due to their effectiveness on ruminant productivity. Therefore, the effect of various inclusion levels of ammonium sulphate on the performance of West African Dwarf (WAD) rams was examined. Sixteen rams weighing 12.8±0.12 kg were assigned to four experimental diets containing 0.00, 0.25, 0.50 and 0.75% levels of ammonium sulphate designated as T1, T2, T3 and T4 respectively. In a completely randomized design, each rams were fed at 5% of their body weight at 60:40 levels of experimental diet and wilted guinea grass as basal diet. The growth trial lasted for 105 days. The chemical composition of the experimental feed and faecal samples were analyzed, and data on total voluntary feed intake, feed conversion ratio, weight gain, nitrogen utilization, and apparent digestibility were also collected. The parameters on proximate composition of the feed were significantly affected by the inclusion of $(NH_4)_2SO_4$ except for DM, GE, Calcium (Ca) and Phosphorus (P). The values for the parameters increased with increased inclusion of $(NH_4)_2SO_4$. The dry matter (DM) and crude protein (CP) obtained ranged from 94.15 - 94.69% and 11.00 - 13.40% respectively while the levels of sulphur was between 4.13 - 4.85%. Live weight gain increased with increased inclusion of $(NH_4)_2SO_4$ and ranged from 3.50 - 5.00 kg but did not differ significantly ($P>0.05$) from each other. The total voluntary feed intake (TVFI) obtained in this study ranged between 59.79 - 62.64 kg and feed conversion ratio (FCR) from 11.96 - 17.90. However, TVFI and FCR decreased with increasing levels of $(NH_4)_2SO_4$ in the diet. Also, acid detergent lignin digestibility was significantly ($P<0.05$) affected by inclusion levels of $(NH_4)_2SO_4$ in the treatments. Nitrogen in feed (3.39g/day), faecal nitrogen output (2.62g/day), urinary nitrogen output (0.21g/day) and nitrogen retention (0.56g/day) were insignificantly ($P>0.05$) difference except percentage nitrogen retained (16.52g/day) which is significantly different ($P<0.05$) from each other. It is evident that rams on 0.75% ammonium sulphate supplementation had better growth performance, improved nutrient digestibility and nitrogen metabolism, hence substantiating its utilization in ram diet.

Keywords: Nutrient intake, growth performance, digestibility, nitrogen metabolism, ammonium sulphate.

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Introduction

Feeds and foods are not equal in their capacity to support the animal functions of maintenance, growth and reproduction (Van Soest, 1994). They supply energy and essential nutrients in the form of protein, vitamins and minerals. Energy and protein are often the most limiting factors for ruminants and have received the most attention in evaluation systems (Mapato *et al.*, 2010). In the formulation of diets for ruminants, it is important to optimize the balance between the energy and protein contents of the feed, so that balanced rumen fermentation occurs and maximum voluntary intake and feed utilization can be achieved (Anusorn *et al.*, 2010).

Dietary protein plays an important role in the nutrition of ruminants, besides providing amino acids; it is also a source of nitrogen for the synthesis of microbial protein (Nocek and Russell, 1988). Therefore, it is considered the most important nutrient and the most expensive, which must be efficiently used. Strategies to reduce the feed cost without interfering negatively in production have been constantly researched. The substitution of traditional conventional feeds in the diets of ruminants is common as economic condition changes (Devendra, 2007).

Rumen micro-organisms living in symbiosis with the host animal require minerals for normal growth and development, among these minerals is sulphur. Sulphur is a necessary component of the amino acids and it represents about 0.25% of the body weight (NRC, 1996). Ammonium sulphate an inorganic form of sulphur contains 24% sulphur and 21% nitrogen. It dissociates easily in the rumen into nitrate and sulphate ions (Mc Donald *et al.*, 1998) while the sulphate ion is reduced in the rumen to suphude ion by a group of bacteria referred to as the sulphur reducing bacteria (Limin-Kung *et al.*, 1998). Ammonium sulphate is relatively inexpensive and freely available (Stephan, 1999) and there is paucity of information on the use of ammonium sulphate as ruminant supplement. Consequently, this study was conducted to determine the effect of ammonium sulphate supplementation on growth performance, nutrient digestibility and nitrogen balance of West African dwarf (WAD) rams.

Materials and Methods

Experimental Site

The study was conducted in sheep unit of the Teaching and Research Farm, University of Ibadan, Nigeria. The location is 70 27'N and 30 45'E at altitude 200-300m above sea level. The climate is humid tropical with mean temperature of 25-290C and the average annual rainfall of about 1250mm.

Animals and Management

Sixteen West African Dwarf (WAD) rams aged between 8-10 months and weighing 12.8 +0.12 kg were randomly allotted to one of four groups with 4 rams per group in a completely randomized design. Each group was assigned to diet containing fixed amounts of urea (NH2)2CO, brewers' dried grain (BDG), palm kernel cake (PKC), dicalcium phosphate (DCP), oyster shell (OS), salt (Nacl), premix (growers) and dry cassava peel with 0.00, 0.25, 0.50 and 0.75% inclusion levels of ammonium sulphate (NH4)2SO4 as T1, T2, T3 and T4 respectively. Prior to the experiment, all the rams were given prophylactic treatment of oxytetracycline L.A. @ 1ml/10kg BW and dewormed with ivomectin at 1ml/10kg BW. Also, the rams were dipped in Amitrax solution to eliminate ecto-parasites. Similarly, Tissue Culture Rinderpest Vaccine (TCRV) was administered against Pestis de Petit Ruminant (PPR) about a week after arrival. The rams were quarantined for a period of 4 weeks and subsequently housed individually in open-sided, well-ventilated pens. All rams had access to fresh water. The rams were fed 5% of their body weight at 60:40 levels of experimental diet and wilted grass between 0800 and 1600hr respectively. The experiment lasted for 105 days. The ingredient composition of experimental diets is presented in Table 1.

Digestibility and Nitrogen Balance Trials

After 105 days growth performance, the rams were transferred into metabolic cages fitted with facilities for separate collection of faeces and urine. The quantity of feed offered, feed refused, faeces and urine were determined after 7 days of adjustment to the cages. About 10% of the faeces and urine collected daily were bulked. Nitrogen loss

from urine and bacteria growth infestation were prevented by introducing 2 drops of concentrated sulphric acid into a well-labeled bottle and stored in a refrigerator. The 10% faeces taken were weighed and used for moisture determination. The remaining faeces were oven dried at 1050C for 24 hours, milled and stored in air tight bottles for chemical composition.

Table 1: Gross Compostion of Experimental Diet (% DM).

Ingredients	T1	T2	T3	T4
Ammonium Sulphate $(NH_4)_2SO_4$	0.00	0.25	0.50	0.75
Urea $(NH_2)_2CO$	1.00	1.00	1.00	1.00
Dry Cassava Peel	60.00	60.00	60.00	60.00
Brewers Dry Grain (BDG)	23.00	23.00	23.00	23.00
Palm Kernel Cake (PKC)	10.00	10.00	1.00	10.00
Dicalcium Phosphate (DCP)	1.00	1.00	1.00	1.00
Oyster Shell (OS)	2.00	2.00	2.00	2.00
Salt (Nacl)	2.00	2.00	2.00	2.00
Premix (Growers)	10.00	1.00	1.00	1.00
Total	**100.00**	**100.25**	**100.50**	**100.75**

T1: 0.00% $(NH_4)_2SO_4$, T2: 0.25% $(NH_4)_2SO_4$, T3: 0.50% $(NH_4)_2SO_4$, T4: 0.75% $(NH_4)_2SO_4$.

Data Collection

The daily feed provided and the leftover of the previous day's feed was weighed to determine the total voluntary feed intake of each ram and was determined on a daily basis.

Feed conversion ratio is the feed intake weight divided by the body weight gain. Samples of experimental diets collected during the experiment were used for proximate analysis. The samples were weighed and dried in an oven at 1050C to constant weight. The dried samples were weighed and ground to pass through a 2mm sieve. The rams' weights were taken once a week before the morning feed was offered.

The milled experimental feed and faecal samples were analyzed for dry matter (DM), crude protein (CP) and ether extract (EE) according to AOAC (1995). The nitrogen content of urine was determined by kjeldahl method according to AOAC (1990) procedure.

Fibre fractions were analyzed by the procedures of Van Soest (1994). Gross energy of feed and faecal samples was determined with bomb calorimeter. Minerals analysis, sulphur and phosphorus of feed and faecal samples were determined using UV spectrometer (AOAC, 2000) while calcium was determined using atomic absorption spectrophotometer (Garcia et al., 2010).

Statistical Analysis

Data generated from parameters investigated were subjected to Analysis of Variance (ANOVA) using Statistical Analysis System (SAS, 1999). Significant differences between treatment means were separated using Least Significant Differences (LSD).

Model of analysis: Completely Randomized Design (CRD) involving 4 treatments and 4 replicates each for sixteen WAD rams (105 days) was conducted.

$Yij=\mu+Tj+\ell ij$

Yij=Individual observation, μ=General mean, Tj=Effect of jth treatment, ℓij= Experimental error.

Results and Discussion

Chemical Composition

Table 2 shows the chemical analysis of experimental diets containing varying level of ammonium sulphate. DM (%) ranged from 94.15 - 94.69 (P>0.05). This implies that, dry matter is an indicator of the amount of nutrient that is available to the animal in a particular feed. The crude protein (CP) of the experimental diets ranged from 13.40% in T4 to 11.00% in T1. The values were within the values of 10-12% recommended by NRC (2000).

Table 2: Chemical Analysis of Experimental Diets Containing Varying Levels of Ammonium Sulphate.

Parameters	T1	T2	T3	T4	Sem
Dry Matter (%)	94.15	94.33	94.51	94.69	0.02
Crude Protein (%)	11.00^{c}	11.80^{c}	12.60^{b}	13.40^{a}	0.02
Crude Fibre (%)	10.51^{c}	11.25^{b}	11.99^{b}	12.73^{a}	0.01
Ether Extract (%)	0.42^{c}	0.72b	1.02^{a}	1.32^{a}	0.01
Ash (%)	16.86^{b}	17.74^{ab}	18.62^{a}	19.50^{a}	0.02
Sulphur (%)	4.13	4.37	4.61	4.85	0.01
Gross Energy (Kcal/kg)	3211.93	3241.53	3271.13	3300.73	0.04
Calcium (%)	0.10	0.10	0.10	0.10	0.003
Phosphorus (%)	0.03	0.03	0.03	0.03	0.002
Ca:P	3:1	3:1	3:1	3:1	0.09

$^{a, b, c}$: Means within rows with unlike superscripts are significantly different from each other ($P<0.05$). T1: 0.00% $(NH_4)_2SO_4$, T2: 0.25% $(NH_4)_2SO_4$, T3: 0.50% $(NH_4)_2SO_4$, T4: 0.75% $(NH_4)_2SO_4$. SEM: Standard Error Mean.

The CP content of the experimental diet is an indication of their nutritional quality since CP content is a very important index of nutritional quality of a feed. This justifies their use as supplements to poor quality crop residues. Thus, ammonium sulphate was added to the treatments to provide adequate sulphur requirement for anaerobic fungi to start the digestion of the components of dietary fibre leading to the production of volatile fatty acids (Okoruwa *et al.,* 2014). Total ash contents were considerably different in values, being highest in T4 (19.50%) and lowest in T1 (16.86%). This implies that, the mineral content present in T4 is higher compared to other treatments in this study. Ether extract had low values in T1 (0.42%), and highest value in T4 (1.32%) and this could be an indication of high level of fat in T4. The gross energy (Kcal) levels were not significantly differed ($P>0.05$) in rams on T4 (3300.73Kcal) compared with T1 (3211.93Kcal).

Growth Performance

Table 3 showed the performance of WAD rams fed ammonium sulphate fortified diet. The feed intake decreases with increasing level of ammonium sulphate. There was significant difference ($P<0.05$) in FCR across the dietary treatments. T1 had the highest FCR (17.90) while T4 (11.96) had the least. The smaller the feed conversion ratio (FCR), the more efficient animals are in converting feed to meat (Smeaton, 2003). Thomas *et al.,* (1951) demonstrated that the addition of inorganic sulphate to a sulphur deficient purified ration improved weight gain and the nitrogen and sulphur retention of sheep. The tendency for a negative effect of T1 on live weight change may be due to reduction in muscular development as a result of depletion of the sulphur-containing amino acids necessary for formation of sulphur-amino acids (Onwuka *et al.,* 1992). Promkot *et al.,* (2007) reported that goats on low sulphur, cassava-based diets had the greatest weight losses as compared to sulphur supplemented groups. It was shown by Ferreiro *et al.,* (1977) that addition of 1g ammonium sulphate per kg of fresh sugar cane improved daily gain significantly on a ration composed otherwise of only sugar cane and urea.

Table 3: Performance of Wad Rams Fed with Diets Fortified with Ammonium Sulphate.

	T1	T2	T3	T4	SEM
Final Body Weight (kg)	18.25	18.75	19.25	19.75	0.01
Live Weight Gain (kg)	3.50	4.00	4.25	5.00	0.01
Daily Weight Gain (kg/day)	0.04	0.04	0.05	0.06	0.002
Total Voluntary Feed-Intake (kg)	62.64	61.69	60.74	59.79	0.01
Daily Feed-Intake (kg/day)	0.70	0.69	0.68	0.66	0.002
Feed conversion Ratio	17.90^{a}	15.42^{b}	13.50^{c}	11.96^{d}	0.02

$^{a, b, c}$: Means within rows with unlike superscripts are significantly different from each other ($P<0.05$). T1: 0.00% $(NH_4)_2SO_4$, T2:0.25% $(NH_4)_2SO_4$, T3:0.50% $(NH_4)_2SO_4$, T4: 0.75% $(NH_4)_2SO_4$. SEM: Standard Error Mean.

Nutrient Digestibility

Table 4 showed the nutrient digestibility in WAD rams fed diets containing varying level of ammonium sulphate. DM digestibility (%) value ranged from 34.15 to 35.68 while CP digestibility (%) value ranged from 36.84 to 38.76. Generally, digestible nutrient intake is a function of apparent nutrient digestibility and dry matter intake. Relative to urea with or without S supplementation in consistent with previous studies Olafadehan *et al.*, (2014) attributed improved intakes of digestible nutrients to greater digestibility of CP of the diet. The EE digestibility (%) values obtained ranged from 30.56 to 32.57. Ash digestibility (%) values for T1 (32.73) to T4 (34.98) were insignificantly ($P>0.05$) differed. Digestible DM and CP intakes showed the same trend as the digestible EE and ash, they are not statistically differed from each other ($P>0.05$). Results revealed significant ($P<0.05$) differences in acid detergent lignin. Enhanced neutral detergent fibre (NDF) digestion of $(NH_4)_2SO_4$ supplemented diets increased dry matter intake (DMI). Increasing the $(NH_4)_2SO_4$ level from 0.00 to 0.75% increased the digestibility of the (NDF) from 42.80 to 43.58%. The result showed that, sulphur helps in the proliferation of fungi and hence they colonize the more rigid lignocellulosic tissues of fibers (Gordon and Philips, 1998).

Table 4: Nutrient Digestibility in Wad Rams Fed with Diets Fortified with Ammonium Sulphate.

	T1	T2	T3	T4	SEM
Dry Matter (%)	34.15	34.66	35.17	35.68	0.02
Crude Protein (%)	36.84	37.48	38.12	38.76	0.02
Crude Fibre (%)	44.67	45.17	45.67	46.17	0.02
Ether Ester (%)	30.56	31.23	31.90	32.57	0.02
Ash (%)	32.73	33.48	34.23	34.98	0.02
Neutral detergent Fibre (%)	42.80	43.06	43.32	43.58	0.02
Acid Detergent Fibre (%)	31.76	32.86	33.96	35.06	0.01
Acid Detergent Lignin (%)	19.67^{d}	22.07^{c}	24.47^{b}	26.87^{a}	0.03

$^{a, b, c}$: Means within rows with unlike superscripts are significantly different from each other ($P<0.05$). T1: 0.00% $(NH_4)_2SO_4$, T2: 0.25% $(NH_4)_2SO_4$, T3: 0.50% $(NH_4)_2SO_4$, T4: 0.75% $(NH_4)_2SO_4$, SEM: Standard Error Mean.

Nitrogen Metabolism

Nitrogen metabolism of rams fed ammonium sulphate supplemented feed and guinea grass is presented in Table 5. Nitrogen (N)-intake for rams on T4 (3.39g/day) was higher and differed from those on T3 (3.27g/day), T2 (3.14g/day), T1 (3.01g/day). This could probably be due to the present of varying level of ammonium sulphate in the experimental diet which improves the utilization of urea as a nitrogen source (Mc Donald *et al.*, 1998). Faecal N-output was also higher in T4 (2.62g/day) although it was insignificantly differing ($P>0.05$) from the value obtained in T1 (2.41g/day) which was lower. This difference observed, might be connected with the degree of sulphur utilization in the treatments. Prvulovic *et al.*, (2012) found that the addition of high protein feed in a diet changes the pattern of nitrogen excretion towards increasing nitrogen excretion in faeces. Urinary N-output values that ranged from 0.18 to 0.21g/day did not varied significantly ($P>0.05$) with rams on T4 having the highest (0.21g/day) value while rams on T1 (0.18g/day) has the lowest.

Table 5: Nitrogen Utilization of Wad Rams Fed with Diets Fortified with Ammonium Sulphate.

Parameter	T1	T2	T3	T4	Sem
Nitrogen Intake (g/day)	3.01	3.14	3.27	3.39	0.04
Feacal Nitrogen (g/day)	24.1	2.48	2.55	2.62	0.04
Urinary Nitrogen (g/day)	0.18	0.19	0.20	0.21	0.02
Nitrogen Output (g/day)	2.59	267	2.75	2.83	0.04
Nitrogen Retained (g/day)	0.42	0.47	0.52	0.56	0.04
% Nitrogen Retained (g/da)	13.95^{b}	14.97^{ab}	15.90^{a}	16.52^{a}	0.04

$^{a, b, c}$: Means within rows with unlike superscripts are significantly different from each other ($P<0.05$). T1: 0.00% $(NH_4)_2SO_4$, T2: 0.25% $(NH_4)_2SO_4$, T3: 0.50% $(NH_4)_2SO_4$, T4: 0.75% $(NH_4)_2SO_4$, SEM: Standard Error Mean.

The insignificant (P>0.05) difference in the amount of faecal N, N intake and quantity of nitrogen retained relative to that consumed or that absorbed among the diets was in support of earlier reports by Bhattacharya *et al.*, (1973) and Chanjula *et al.*, (2008). It is now well established that nitrogen retention depends on the intake of nitrogen in the diet Sarwar *et al.*, (2003). Higher (P<0.05) urinary N excretion in urea diet could be due to rapid degradation of urea by rumen microbes to produce excess NH_3-N which is absorbed and excreted in the urine in the form of urea. Souza *et al.*, (2009) reported that bucks fed urea-supplemented diets excreted more urinary N than those fed natural proteins. It appears that urea supplementation with S helps in improving urea metabolism in the rumen which possibly accounts for reduction in urinary N excretion, Olafadehan *et al.*, (2014).

N-balance and retention which were function of nitrogen ingested and digested (Okoruwa *et al.*, 2013) was not significantly differed (P>0.05) with T4 (0.56g/day) recorded the highest, while T1 recorded the lowest (0.42g/day). The low N-balance and retention observed in T1 might be influenced by the absence of ammonium sulphate in the treatment. However, the study treatments posted the best positive N-balance for T4, suggesting that the protein requirements for the rams to improve rumen microbial activity were adequately met by the diets. This confirmed the report of (Nocek *et al.*, 1988) who found that rams will live and grow quite normally on a treatment that is having up to 0.60% sulphur. This is true because inclusion of ammonium sulphate at 0.75% performed better than other treatments.

Conclusion

Ammonium sulphate is a chemical compound that serves as a ready source of nitrogen and sulphur, which could play an important role in enhancing the growth performance of the WAD rams. The ease of preparation and maintenance make the experimental feed easy for adoption by small-scale farmers at rural level. Ration fortified with ammonium sulphate offered a balance of essential nutrients, which is the major determinant of dry matter intake and growth rate and so would overcome dry season weight losses and rather poor performance in sheep fed cut and carry fodder.

It was observed that diets fortified with ammonium sulphate at 0.75% were associated with optimum performance and considerable improvement in. This is because diets supplemented with ammonium sulphate at 0.75% could be associated with optimum performance and considerable improvements in:

Digestibility of nutrients,
Nutritive values of digestible portions,
Nitrogen-sulphur utilization,
Maximization nutrients from the rumen and
Manipulation of rumen function to increase nutrient capture.

Therefore, ammonium sulphate fortification at 0.75% in diet formulation for West African dwarf sheep could be recommended without any harmful effect.

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